Claims

1. Method for treating a crystal with a solution containing one ore more molecule species, wherein the molecules have a molecular weight of < 500 Da, comprising the following steps:
   - the crystal is fixed on a holding device, in particular without being embedded in a liquid environment, and
   - microdrops of the liquid are applied onto the crystal.

2. Method for treating a crystal according to claim 1, wherein the molecules contained in the solution have a molecular weight of < 200 Da.

3. Method for treating a crystal according to claim 1 or 2, wherein the molecules contained in the solution have a molecular weight of < 100 Da.

4. Method for treating a crystal according to any one of the preceding claims, wherein the crystal is a protein crystal.

5. Method for treating a crystal according to any one of the preceding claims, wherein the molecules contained in the solution bind to the proteins in the protein crystal as ligands, preferably with an affinity between 10^{-3} and 10^{-4} M.

6. Method for treating a crystal according to any one of the preceding claims, wherein the molecules contained in the solution or the molecules of at least one molecule species contained in the solution have at least one electron-rich or anomalous dispersion center, preferably a heavy(metal)atom.

7. Method according to any one of the preceding claims, wherein furthermore a defined environment is generated around the crystal during the application of microdrops onto the crystal.

8. Method according to claim 7, wherein generating a defined environment comprises generating a gas stream of defined composition around the crystal.

9. Method according to claim 8, wherein the gas stream consists of an air stream with controlled air humidity.

10. Method according to claim 8, wherein the gas stream is regulated during the drip-on procedure.

11. Method according to any one of claims 9 or 10, wherein the air humidity of the gas stream and the frequency, at which the drops are dripped onto the crystal by means of the micro dosage system, are synchronized during the drip-on procedure in such a way that the crystal is strained as little as possible and, in particular, that the volume of the crystal alters by no more than 20%, in particular by no more than 10%.

12. Method according to any one of claims 8 to 11, wherein the gas stream comprises a solubilizer at a controlled concentration for a substance to be applied onto the crystal.

13. Method according to any one of claims 1 to 12, wherein the volume of the microdrops is smaller than the volume of the crystal.

14. Method according to claim 13, wherein the microdrops of the solution have a volume of between 1 nl and 100 pl, preferably between 100 pl and 20 pl, and also preferably between 20 pl and 4 pl.

15. Method according to any one of the preceding claims 1 to 14, wherein the solution containing molecule species and applied onto the crystal is an aqueous solution or a solution, at least partially, comprising organic solvents and, optionally, being heated up to more than 20°C.

16. Method according to claim 15, wherein solution containing the molecule species consists of or contains a volatile organic solvent.

17. Method according to claim 16, wherein the solvent consists of or contains DMSO.

18. Method according to claim 15 or 16, wherein the solvent containing the molecule species is or contains a preferably entirely volatile organic solvent, which boils at a temperature of below 100°C.

19. Method according to any one of claims 15 to 18, wherein the solvent contains DMSO, trifluoroethanol, acetone, chloroform, and/or methanol.

20. Method according to any one of the preceding claims, wherein the molecules contained in the solution to be applied onto the crystal are hardly water-soluble.

21. Method according to any one of the preceding claims, wherein the solution contains a cocktail of at least 3, more preferably at least 10, even more preferably at least 20, and most preferably at least 50 different molecule species.

22. Method according to any one of the preceding claims, wherein the solution contains at least one molecule species at a concentration of 10^{-1} to 10^{-3} M.

23. Method according to any one of the preceding claims, wherein a method step is inserted before the method, by means of which fragments potentially binding to a target structure are identified, in particular a method step, which is based on a spectroscopic method, for example NMR spectroscopy or surface plasmon resonance spectroscopy, or an in silico docking method.

24. Method according to any one of claims 1 to 23, wherein the gas stream contains one or more substance/s, which contain/s one or more ligand/s and/or inhibitor/s.

25. Method for determining a crystallographic structure of a complex of, for example, a protein and at least one molecule species, wherein (a) the method steps according to any one of claims 1 to 24 are conducted, (b) the crystal is irradiated with X-ray or synchrotron radiation, and (c) the diffraction image of the crystal is recorded.

26. Method for determining a crystallographic structure according to claim 25, wherein (d) an electron density map is calculated by means of using the phase information and the intensity of the reflexes in the diffraction image and the binding site and positioning of the at least one bound molecule species is determined, for example, in the protein structure.

27. Method according to claim 26, wherein the phase information is obtained by means of the use of heavy metal atom derivatives ("isomorphous replacement"), "molecular replacement", or MAD (multiple anomalous scattering).

28. Method for determining a crystallographic structure according to claim 26 or 27, wherein the binding site and positioning of the at least one bound molecule species in the, for example, protein structure is determined from the difference of electron densities of non-complexed and complexed structure by means of a electron density difference map.

29. Method according to any one of claims 25 to 27, wherein the irradiation is conducted with monochromatic X-ray radiation or with synchrotron radiation during the treatment of the crystal with the solution.

30. Method for identifying molecules binding a crystallized protein, wherein (a) at least one molecule species is applied onto the crystal according to a method according to any one of claims 1 to 24, (b) diffraction intensities are measured at intervals of variable length, and (c) said diffraction intensities measured at intervals are compared with respect to their time-dependent sequence.

31. Method for identifying of a ligand binding the target structure, wherein (a) a method according to any one of claims 1 to 24 is conducted, (b) the structure of at least one complex having at least two fragments is determined according to a method according to any one of claims 25 to 29, (c) linker/s to a ligand, which is/are located between the at least two fragments, is/are determined, and (d) a ligand containing the at least two fragments and the at least one linker is synthesized.

32. Method according to any one of claims 1 to 24, wherein the method is conducted by means of using a device for treating a crystal with a substance having a holder for fixing the crystal and at least one micro dosage system, which is arranged in relation to the holder in such a way that it can apply microdrops of the liquid onto the crystal fixed in the holder.

33. Method according to claim 32, wherein the device used according to the method furthermore comprises a device, by means of which a defined environment can be generated around the crystal during the drip-on procedure.

34. Method according to any one of claims 32 or 33, wherein the device allows the generation of a defined environment by means of generating a gas stream of defined composition around the crystal.

35. Method according to claim 32 or 34, wherein furthermore the holder is developed in such a way that the gas stream can be led through the holder in such a way that it is directed toward the crystal fixed in the holder.

36. Method according to any one of the preceding claims, wherein a device having a holder consisting of a carrier block for a holder capillary, which has a free support end for the crystal, is used.

37. Method according to claim 36, wherein a device having a holder capillary consisting of a micropipette, in which a negative pressure can be generated in order to hold the crystal, is used.

38. Method according to any one of claims 36 or 37, wherein the carrier block of the holder of the device used in accordance with the present invention has an integrated gas channel having a mouth end, which is directed toward the support end of the holder capillary.

39. Method according to any one of claims 34 to 38, wherein a device is used, which furthermore has a gas mixing device, by means of which the composition of the gas stream can be variably adjusted.

40. Method according to claim 39, wherein a device is used, in which the gas consists of air having a specific humidity content and the gas mixing device is developed in such a way that by means of it the air humidity can be adjusted.

41. Method according to any one of claims 34 to 40, wherein a device is used, which furthermore comprises a device for adding a solubilizer, by means of which a solubilizer for a substance to be introduced into the crystal structure of the crystal can be added to the gas stream.

42. Method according to claim 41, wherein a device is used, which furthermore comprises a concentration adjusting device for adjusting the concentration of the solubilizer.

43. Method according to any one of claims 34 to 42, wherein a device is used, which furthermore comprises a temperature regulating device, by means of which the temperature of the gas stream can be variably adjusted.

44. Method according to any one of the preceding claims, wherein a device is used, in which the micro dosage system is developed in such a way that it can generate microdrops of the liquid to be applied onto the crystal, which have a volume that is smaller than the volume of the crystal.

45. Method according to claim 44, wherein a device is used, in which the micro dosage system is developed in such a way that it can generate microdrops having a volume of between 10 and 20 percent of the volume of the crystal and preferably between 5 and 10 percent of the volume of the crystal.

46. Method according to any one of claims 42 or 43, wherein the micro dosage system is developed in such a way that it can generate microdrops having a volume of between 1 nl and 100 pl, preferably between 100 pl and 20 pl, and also preferably between 20 pl and 4 pl.

47. Method according to any one of the preceding claims, wherein a device is used, in which the micro dosage system furthermore has a liquid supply system, by means of which different liquids to be dripped onto the crystal can be supplied to a drop generating part of the micro dosage system in a time-dependently controlled manner.

48. Method according to claim 47, wherein a device is used, in which the liquid supply system of the micro dosage system comprises an electrically controllable precision syringe and a duct system, with which the precision syringe can be connected, via electrically controllable valves, with different liquid supply containers and with the drop generating part of the micro dosage system in order to supply liquid for drop generation to the latter.

49. Method according to any one of the preceding claims, wherein a device is used, in which the micro dosage system is developed in such a way that it comprises a piezo pipette, which forms the drop generating part.

50. Method according to any one of the preceding claims, wherein the crystal is vapor-plated with solvent, in particular with organic solvent, by means of an evaporator.

51. Method for X-ray crystallographic structure determination at high throughput, wherein (a) the crystal/s is/are held ready, preferably in a freely mounted manner, (b) microdrops of a solution containing, for example, at least one ligand are applied onto the preferably freely mounted crystals, (c) the crystals treated according to method step (b) are stored, and (d) the crystals are examined X-ray crystallographically.